PATENT COOPERATION TREATY

From the
INTERNATIONAL SEARCHING AUTHORITY

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PCT

WRITTEN OPINION OF THE
INTERNATIONAL SEARCHING AUTHORITY

(PCT Rule 43*bis*.1)

Date of mailing *(day/month/year)* 25 SEP 2007

Applicant's or agent's file reference
UCIVN-061pc

FOR FURTHER ACTION
See paragraph 2 below

International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/US 05/10266	29 March 2005 (29.03.2005)	29 March 2004 (29.03.2004)

International Patent Classification (IPC) or both national classification and IPC
IPC(8) - C12Q 1 / 68 (2007.01)
USPC - 435/6

Applicant The Regents of University of California

1. This opinion contains indications relating to the following items:

- ☒ Box No. I Basis of the opinion
- ☐ Box No. II Priority
- ☐ Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
- ☐ Box No. IV Lack of unity of invention
- ☒ Box No. V Reasoned statement under Rule 43*bis*.1(a)(i) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
- ☐ Box No. VI Certain documents cited
- ☐ Box No. VII Certain defects in the international application
- ☐ Box No. VIII Certain observations on the international application

2. **FURTHER ACTION**

If a demand for international preliminary examination is made, this opinion will be considered to be a written opinion of the International Preliminary Examining Authority ("IPEA") except that this does not apply where the applicant chooses an Authority other than this one to be the IPEA and the chosen IPEA has notified the International Bureau under Rule 66.1*bis*(b) that written opinions of this International Searching Authority will not be so considered.

If this opinion is, as provided above, considered to be a written opinion of the IPEA, the applicant is invited to submit to the IPEA a written reply together, where appropriate, with amendments, before the expiration of 3 months from the date of mailing of Form PCT/ISA/220 or before the expiration of 22 months from the priority date, whichever expires later.

For further options, see Form PCT/ISA/220.

3. For further details, see notes to Form PCT/ISA/220.

Name and mailing address of the ISA/US	Date of completion of this opinion	Authorized officer
Mail Stop PCT, Attn: ISA/US Commissioner for Patents P.O. Box 1450, Alexandria, Virginia 22313-1450 Facsimile No. 571-273-3201	12 April 2007 (12.04.2007)	Lee W. Young PCT Helpdesk: 571-272-4300 PCT OSP: 571-272-7774

Form PCT/ISA/237 (cover sheet) (April 2005)

WRITTEN OPINION OF THE INTERNATIONAL SEARCHING AUTHORITY

International application No.
PCT/US 05/10266

Box No. I Basis of this opinion

1. With regard to the **language**, this opinion has been established on the basis of:

   ☒ the international application in the language in which it was filed

   ☐ a translation of the international application into _____ , which is the language of a translation furnished for the purposes of international search (Rules 12.3(a) and 23.1(b)).

2. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application and necessary to the claimed invention, this opinion has been established on the basis of:

   a. type of material

      ☐ a sequence listing

      ☐ table(s) related to the sequence listing

   b. format of material

      ☐ on paper

      ☐ in electronic form

   c. time of filing/furnishing

      ☐ contained in the international application as filed

      ☐ filed together with the international application in electronic form

      ☐ furnished subsequently to this Authority for the purposes of search

3. ☐ In addition, in the case that more than one version or copy of a sequence listing and/or table(s) relating thereto has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

4. Additional comments:

WRITTEN OPINION OF THE INTERNATIONAL SEARCHING AUTHORITY

International application No.
PCT/US 05/10266

Box No. V **Reasoned statement under Rule 43*bis*.1(a)(i) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement**

1. Statement

Novelty (N)	Claims	1-34	YES
	Claims	NONE	NO
Inventive step (IS)	Claims	NONE	YES
	Claims	1-34	NO
Industrial applicability (IA)	Claims	1-34	YES
	Claims	NONE	NO

2. Citations and explanations:

Claims 1-34 lack an inventive step under PCT Article 33(3) as being obvious over US 2003/0087858 A1 (HERRNSTADT).

As per claim 1, Hermstadt discloses a method for diagnosis of Alzheimer's disease by detecting genetic mutations in mitochondrial cytochrome c oxidase ("CR") genes and suppressing these mutations or their effects in the treatment of Alzheimer's disease (para [0002]). It would have been obvious for a person having ordinary skills in the art how to detect mtDNA CR mutations and how to use them for diagnosing a disorder associated with the development of beta amyloid deposits or fibrils for the following reasons: 1) because it is well known in the art that mtDNA CR is the primary site for the regulation of mtDNA transcription and replication, 2) that Alzheimer's Disease is a progressive neurodegenerative disease that is associated with the accumulation of 13-amyloid (A13) plaques and neuritic tangles in the brain.

As per claim 2, directed to the method of claim 1, further comprising making a qualitative determination that mtDNA CR mutation is or is not present, it is obvious for reasons set forth for claim 1, and further because it would have been obvious to one of ordinary skill in the art how to detect the exists of genetic mutations of mtDNA CR region.

As per claim 3, directed to the method of claim 1, further comprising quantitative determination of mtDNA CR mutations, it is obvious for reasons set forth for claim 1, and further because Herrnstadt discloses use of probes for quantitative analysis of wild-type and mutant mtDNA samples (para [0156]).

As per claims 4 and 5, directed to the method of claim 3, further comprising the step of comparing a mtDNA CR value to either a control mtDNA CR value or a mtDNA CR value representative of subjects who suffer from a disorder, respectively, they are obvious for reasons set forth for claim 3, and further because having negative or positive control is the basic requirement for a meaningful scientific experiment.

As per claim 13, directed to the method of claim 1, wherein Step A is carried out at least in part by PNA-clamping PCR, it is obvious for reasons set forth for claim 1, and further because Hermstadt discloses use of the polymerase chain reaction for detecting the specific mutations in the mitochondrial genes (para [0062]). It would have been obvious for a person having ordinary skills in the art the advantages of using PNA-clamping PCR to detect the mtDNA CR mutations because it is well known in the art that PNA-clamping PCR can avoid problems with contamination by combining amplification and detection in a closed system.

As per claims 14 and 15, directed to the method of claim 1, wherein Step A is carried out at least in part by oligonucleotide hybridization or primer extension, respectively, they are obvious for reasons set forth for claim 1, and further because Herrnstadt discloses the use of single nucleotide primer-guided extension assays or hybridization techniques using target-specific oligonucleotides for detecting the specific mutations in the mitochondrial genes (para [0062]).

As per claim 16, directed to the method of claim 1, wherein Step A is carried out at least in part by restriction digestion, it is obvious for reasons set forth for claim 1, and further because it would have been obvious to one of ordinary skill in the art to include restriction digestion into the method.

As per claim 17, directed to the method of claim 1, wherein Step A is made in a group of specific specimen of tissue, cells or body fluid, it is obvious for reasons set forth for claim 1, and further because Herrnstadt discloses to harvest DNA from blood and brain samples (para [0040]).

As per claims 18 and 19, directed to the method of claim 1, wherein the method is carried out for post-symptomatic or pre-symptomatic diagnosis of a disorder, they are obvious for reasons set forth for claim 1, and further because Herrnstadt discloses methods for detecting such mutations as a diagnostic for Alzheimer's disease, either before or after the onset of clinical symptoms (para [0013]).

--Please See Continuation Sheet--

WRITTEN OPINION OF THE
INTERNATIONAL SEARCHING AUTHORITY

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Supplemental Box

In case the space in any of the preceding boxes is not sufficient.
Continuation of:

As per claims 20 and 21, directed to the method of claim 1, they are obvious for reasons set forth for claim 1, and further because Hermstadt discloses that degenerative diseases such as Leber's hereditary optic neuropathy, myoclonus, epilepsy, lactic acidosis and stroke (MELAS), and myoclonic epilepsy ragged red fiber syndrome are transmitted through mitochondrial DNA mutations and the methods can be used to detect such mutations as a diagnostic for Alzheimer's disease (para [0007]).

As per claims 22-29, directed to the method of claim 1, they are obvious for reasons set forth for claim 1, and further because it is well known in the art that amyloid deposits or fibrils are thought to be involved in the pathogenesis of various amyloid diseases of genetic, infectious and/or spontaneous origin, including but not limited to Alzheimer's disease (AD), spongiform encephalopathies, Parkinson's disease, type II diabetes, Creutzfeldt-Jakob disease, Down's Syndrome-associated dementia, Huntington's disease, macular degeneration,various prion diseases and numerous others.

As per claim 30, directed to the method of claim 1, it is obvious for reasons set forth for claim 1, 13 and 17, and further because Hermstadt discloses that cloning and sequencing of the COX genes can serve to detect AD mutations in patient samples (para [0063]). It would have been obvious to one of ordinary skill in the art to obtain sample cells, extract DNA, amplify mtDNA CR, and detect the 414 and 477 nucleotide variants by sequencing and then cloning the mutant molecules and sequencing the clone.

As per claim 31, directed to the method of claim 1, it is obvious for reasons set forth for claim 1, and further because one of ordinary skill in the art would have known how to select appropriate reagents and/or materials necessary for detection of the mtDNA CR mutation.

As per claim 32, directed to the method of claim 31, it is obvious for reasons set forth for claim 31, and further because instructions for use of the test system do not rise to to the level of a patentable advance.

As per claim 33, directed to the method of claim 31, it is obvious for reasons set forth for claim 31, and further because it is well known in the art to include the standard control data as the reference.

As per claim 34, directed to the method of claim 33, it is obvious for reasons set forth for claim 31, and further because it is well known in the art to use computer software to provide the reference.

Claims 6-12 lack an inventive step under PCT Article 33(3) as being obvious over HERRNSTADT in view of US 6462190 B1 to Michikawa et al. (hereinafter "MICHIKAWA").

As per claims 6,7,9,10 and 12, directed to the method of claim 1, further comprising testing for T414G, T414C, T146C, T152C, T195C mutations, they are obvious for reasons set forth for claim 1, and further because Michikawa discloses a method for detecting the presence or risk of age-related disorders by determining the presence of at least one mutation in a mitochondrial DNA main control region including positions 1 and 660 of the Cambridge sequence (col1 ln55- col2 ln5) and mutations are selected from the group consisting of T414G, A368G , T285C, A249G, T195C, T152C, T146C (col2 ln25-35), T414C (col4 ln15-20).

As per claim 8, directed to the method of claim 1, further comprising testing for T477C mutation, it is obvious for reasons set forth for claim, and further because the mutation is in the control region of mt DNA, the primary site for the regulation of mtDNA transcription and replication. It would have been obvious to one of ordinary skill in the art that the T477C mutation may have the same effect as the T414C mutation.

As per claim 11, directed to the method of claim 1, further comprising testing for A189G mutation, it is obvious for reasons set forth for claim 1 and 8, and further because it would have been obvious to one of skill in the art that the mutation A189G may have the same effect as the A249G mutation.

Claims 1-34 have industrial applicability as defined by PCT Article 33(4) because the subject matter can be made or used in industry

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WO 2007/011322
PCT/US2005/010266

From the INTERNATIONAL BUREAU

PCT

NOTIFICATION CONCERNING
SUBMISSION OR TRANSMITTAL
OF PRIORITY DOCUMENT

(PCT Administrative Instructions, Section 411)

To:

BUYAN, Robert, D.
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Date of mailing *(day/month/year)* 05 February 2007 (05.02.2007)	
Applicant's or agent's file reference UCIVN-061pc	IMPORTANT NOTIFICATION
International application No. PCT/US2005/010266	International filing date *(day/month/year)* 29 March 2005 (29.03.2005)
International publication date *(day/month/year)* 25 January 2007 (25.01.2007)	Priority date *(day/month/year)* 29 March 2004 (29.03.2004)
Applicant THE REGENTS OF THE UNIVERSITY OF CALIFORNIA et al	

1. By means of this Form, which replaces any previously issued notification concerning submission or transmittal of priority documents, the applicant is hereby notified of the date of receipt by the International Bureau of the priority document(s) relating to all earlier application(s) whose priority is claimed. Unless otherwise indicated by the letters "NR", in the right-hand column or by an asterisk appearing next to a date of receipt, the priority document concerned was submitted or transmitted to the International Bureau in compliance with Rule 17.1(a) or (b).

2. *(If applicable)* The **letters "NR"** appearing in the right-hand column denote **a priority document which, on the date of mailing of this Form, had not yet been received by the International Bureau** under Rule 17.1(a) or (b). Where, under Rule 17.1(a), the priority document must be submitted by the applicant to the receiving Office or the International Bureau, but the applicant fails to submit the priority document within the applicable time limit under that Rule, **the attention of the applicant is directed** to Rule 17.1(c) which provides that no designated Office may disregard the priority claim concerned before giving the applicant an opportunity, upon entry into the national phase, to furnish the priority document within a time limit which is reasonable under the circumstances.

3. *(If applicable)***An asterisk (*)** appearing next to a date of receipt, in the right-hand column, denotes **a priority document submitted or transmitted to the International Bureau but not in compliance with Rule 17.1(a) or (b)** (the priority document was received after the time limit prescribed in Rule 17.1(a) or the request to prepare and transmit the priority document was submitted to the receiving Office after the applicable time limit under Rule 17.1(b)). Even though the priority document was not furnished in compliance with Rule 17.1(a) or (b), the International Bureau will nevertheless transmit a copy of the document to the designated Offices, for their consideration. In case such a copy is not accepted by the designated Office as the priority document, Rule 17.1(c) provides that no designated Office may disregard the priority claim concerned before giving the applicant an opportunity, upon entry into the national phase, to furnish the priority document within a time limit which is reasonable under the circumstances.

Priority date	Priority application No.	Country or regional Office or PCT receiving Office	Date of receipt of priority document
29 March 2004 (29.03.2004)	60/557,612	US	09 November 2006 (09.11.2006)

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